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(54) Title: MULTI-FLUORESCENT HAIRPIN ENERGY TRANSFER OLIGONUCLEOTIDES (57) Abstract <p>An oligonucleotide, labeled with a molecular energy transfer trio and containing two sequences capable of hairpin formation, is used in the detection of two targets by irradiation with a single wavelength of light. One of the two sequences contains an energy donor and a first energy acceptor, and the other sequence contains a second energy acceptor. The donor is in close proximity to the second acceptor only if the hairpin is formed, while the donor is always in close proximity to first acceptor. A sample is assayed, using this oligonucleotide in conjunction with another oligonucleotide which contains the donor fluorophore and the quencher, arranged as described above, but which lacks the acceptor fluorophore. The present oligonucleotide and the other oligonucleotide are specific to first and second targets, respectively. If a sample contains the first and second targets, then hairpin formation is prevented after each oligonucleotide is incorporated into a target amplification product or hybridized to a target. Subsequent irradiation of the sample with the single wavelength of light which excites the donor fluorophore, but not the acceptor fluorophore, causes two distinctive signals to be generated. The first signal is emitted by the second fluorophore of the present oligonucleotide, while the second signal is emitted by the first fluorophore of the other oligonucleotide. Thus, the first and second targets are detected when one observes the first and second signals, respectively.</p>		